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INTRA-VITAM STAINING OF TUBERCULOUS GUINEA-PIGS WITH FAT-SOLUBLE DYES

(SUPPLEMENTARY NOTE)*

STUDIES ON THE BIOCHEMISTRY AND CHEMOTHERAPY OF TUBERCULOSIS. VI.

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In a preceding article of this series,¹ I reported the results of a series of experiments directed to determining the effects of fat-soluble dyes administered to guinea-pigs, both tuberculous and normal. Since the publication of this paper, I have completed experiments with other dyes not previously considered, and although the results are much the same, it seems desirable to report them briefly for the sake of completeness. Attention is also called to the summary of experiments performed on the effect of fat stains upon the growth of tubercle bacilli in artificial media, which is incorporated in the immediately preceding paper of Miss Sherman.

FEEDING OF FAT STAINS TO TUBERCULOUS GUINEA-PIGS.

Indulin (fat-soluble).—Four guinea-pigs were inoculated with tubercle bacilli and fed a 1 per cent suspension of indulin, Grüber, in peanut oil, 2–4 c.c. every second day, during the entire period of infection, being killed on the 65th day after infection. The organs—tuberculous lymph glands, spleen, kidneys, liver, adrenals, brain, fatty tissues, and lungs—were removed, ground up with anhydrous sodium sulfate, dried, and extracted in a Greene apparatus with alcohol.² None of the organs or tuberculous areas revealed any of the blue stain except the lungs, from which the alcohol extract was a distinct blue or bluish green, and section revealed stained fat in the air passages. This was easily accounted for by aspiration of the oil, containing the dye.

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¹ *Jour. Infect. Dis.*, 1912, 11, p. 373.

² The indulin used was soluble in alcohol and CHCl₃, but practically not at all in ether, petroleum ether, and water.

Dimethylaminoazobenzol.—Four guinea-pigs were inoculated with tuberculosis and fed a 1 per cent solution of dimethylaminoazobenzol (Grübler) in peanut oil, 2–4 c.c. every other day, during the entire period of infection, 65 days, at the end of which time the tissues were studied microscopically (frozen sections) and chemically, as above, grinding them with anhydrous sodium sulfate and extracting with ether.¹ On account of the fact that most of the organs and fats of the body gave a yellow ether extract, no definite conclusions could be reached from the chemical examination, but, microscopically and macroscopically, none of the organs or tuberculous areas revealed definite yellow staining.

Bismarck brown.—Five guinea-pigs were inoculated with tubercle bacilli and four of these fed Bismarck brown, 1 per cent suspension, in oil, and one a 1 per cent watery solution of 2–4 c.c., every second day, the experiment lasting 65 days, at the end of which time the organs and tuberculous tissues were examined histologically and chemically by extraction² so far as possible. Most organs normally gave a brown extract with alcohol. As far as could be judged from the examination none of the organs or tuberculous areas had been stained.

Negative results were also obtained in short-period experiments of 14 days with alkanin (Fettlosl. Roth), Grübler, and Annatto (1 per cent in oil *per os*). Examinations were only histological.

Summary.—Indulin, dimethylaminoazobenzol, (1 per cent in oil), and Bismarck brown, (1 per cent in oil and in water), fed to tuberculous guinea-pigs, do not appear to enter the organs nor tuberculous areas to any appreciable extent when given for a period of about 65 days. Alkanin and annatto, 1 per cent in oil, do not enter within a period of 14 days.

¹ The dimethylaminoazobenzol used was readily soluble in ether, CHCl_3 , petroleum ether, and alcohol, but insoluble in water.

² The Bismarck brown used was soluble in water, alcohol, and petroleum ether, but insoluble in CHCl_3 and ether.